

in this study elicited strong IFN- $\gamma$  and TNF- $\alpha$  responses.

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# **Prevalence of genes responsible for resistance to antimicrobials in surface water *Escherichia coli* isolates**

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**Background:** The resistance to antimicrobial agents is an important issue in both human and veterinary medicine. The excessive use of antibiotics is hastening the development of antibiotic resistance in bacteria augment health risks to humans and animals. Lack of access to potable water has forced many inhabitants in developing countries to rely on surface water resources for their daily water needs. Presently, these water resources have emerged as reservoirs of *Escherichia coli* pathotypes harboring virulence as well as multi-drug resistant genes which could play an important role in the diarrheal disease outbreaks. The river Ganga and its tributaries meet 40% of the water requirement for drinking and irrigation in India.

**Methods:** In this study, *E. coli* isolates (n=65) retrieved from the river Ganga and Gomti (a major tributary of the river Ganga) were screened using Polymerase Chain Reaction for prevalence of genes (*bla*TEM, *bla*SHV, *aac*(3)-IIa, *aac*(3)-IV, *aph*(3')-Ia, *aph*(3')-IIa, *ant*(3'')-Ia (*aad*AI), *ant*(3'')-If (*aad*A6), *tet*A, *tet*B, *tet*C, *cat*I, *flo*R, *sul*1, *sul*2) responsible for resistance to antimicrobial agents of five antimicrobial families ( $\beta$ -lactams, Aminoglycosides, Tetracycline, Phenolics, Sulfonamides).

**Results:** Our observations indicate that 67.2, 32.3, 55.3, 72.3 76.9, 63.0, 75.3, 43.0, 44.6% *E. coli* isolates exhibit *tet*A, *tet*B, *tet*C, *bla*TEM, *bla*SHV, *cat*I, *flo*R, *sul*1, *sul*2 genes, respectively.

**Conclusion:** The prevalence of *E. coli* isolates harboring multiple antimicrobial resistance genes points to the inherent health risks associated with the use of surface water by inhabitants of the planned and temporary settlements along the banks of these rivers. This will require formulation of strategies for preemptive monitoring of surface water to prevent diarrheal outbreaks.

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# **E gene variation with reference to neurovirulence in the Indian clinical isolates of Japanese encephalitis virus**

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**Background:** Japanese encephalitis (JE) is an important arboviral infection of public health concern. There is a significant variation in mortality (20-60%) in JE viral infection. It is possibly attributed due to the host genetic makeup or genomic variation in the JE virus. Various approaches have allowed E gene sequences of flaviviruses to be related to virulence in animal models and shows single amino acid substitutions are sufficient to alter neurovirulence and neuroinvasiveness. The present study has looked for the mutational analysis of E gene, in clinical isolates at amino acid positions at 176,177,227,244,264 and 279, which have been shown responsible for neuro-virulence in experimental animals.

**Methods:** A total of 95 patients with suspected viral encephalitis were enrolled. JEV conformation was done by MAC ELISA and RT-PCR. The RT-PCR positive samples were further subjected to sequencing using ABI PRISM BigDye Terminator cycle sequencing ready reaction kit in ABI PRISM 310 genetic analyzer. The drafting of sequences was performed using BioEdit software. Neighbor joining algorithm was implemented for phylogenetic inference using MEGA 4.0.2. The DNA sequences were translated insilico and mutation analysis was performed. Re-confirmation of mutations was done using BLAST tool in NCBI website

**Results:** Among confirmed cases 70% belonged to the pediatric age group, with a male to female ratio of 3:1. Patients presented with moderate to high-grade fever (41%); convulsions and rigidity (65%), extra pyramidal features (35%). Convulsion was often the presenting symptom. A mortality of 27% was observed among JE positive cases. JE virus specific RNA was detected in 7 cases. Phylogenetically all our isolates belonged to genotype-III. Interestingly a novel mutation of S227T at amino acid level was detected corresponding to the domain II of E gene in JEV compared to both Indian and overseas isolates.

**Conclusion:** Genotype III was found to be circulating in this part of India. With the present available limited number of cases no significant correlation was found between E gene mutation and disease severity. However, the observation of novel mutation S227T of E protein in this geographical area has given the impetus to explore its role in JE pathogenesis and vector competency

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